MATLAB code used in Kobayashi & Matsuo, *Cell Reports* (2023)

1. License
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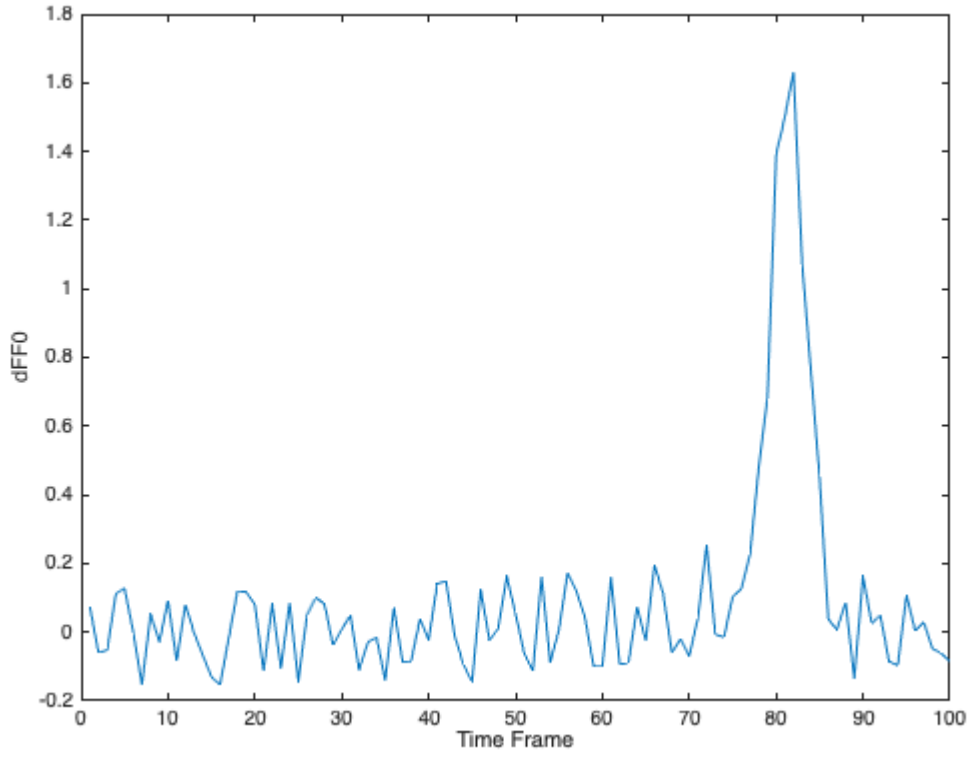
Generate DEMO data

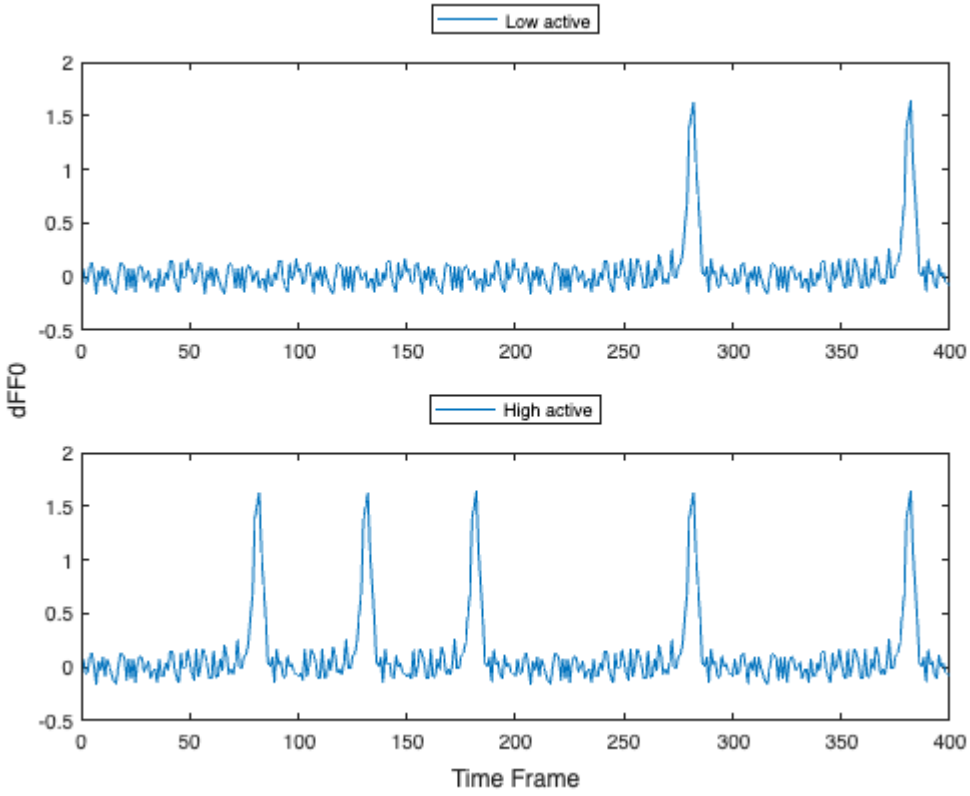
1. dFF0_data: 1600 frames-by-30 cells matrix of ΔF / F0 data .
2. Session_labels: 4-by-1 string vector; "ContextA1","ContextA2","ContextB1", and "ContextB2".
3. Frame_idx: 1600-by-1 string vector; "ContextA1","ContextA2","ContextB1", or "ContextB2".

```
recording_Hz = 20;

num_of_recording_frames_of_a_session = 400; % 20/400 = 20 sec
num_of_recording_cells = 30;

dFF0_data=zeros(num_of_recording_frames_of_a_session*4,num_of_recording_cells);
Session_labels = ["ContextA1";"ContextA2";"ContextB1";"ContextB2"];
Frame_idx = [repmat(Session_labels(1),num_of_recording_frames_of_a_session,1);...
             repmat(Session_labels(2),num_of_recording_frames_of_a_session,1);...
             repmat(Session_labels(3),num_of_recording_frames_of_a_session,1);...
             repmat(Session_labels(4),num_of_recording_frames_of_a_session,1)];


[~, y0] = titanium; %1-by-49 vector

rng default % for reproducibily
y1 = [y0,0.6]; %1-by-50 vector
y2 = y1-min(y1); %1-by-50 vector, represents a calcium event
one_event_trace = y2+(0.5-rand(size(y2)))/3; %1-by-50 vector with noise
no_event_trace = (0.5-rand(size(y2)))/3; %1-by-50 vector, bakcground fluctuation

figure; plot([no_event_trace,one_event_trace]);
ylabel ("dFF0");
xlabel("Time Frame");
```



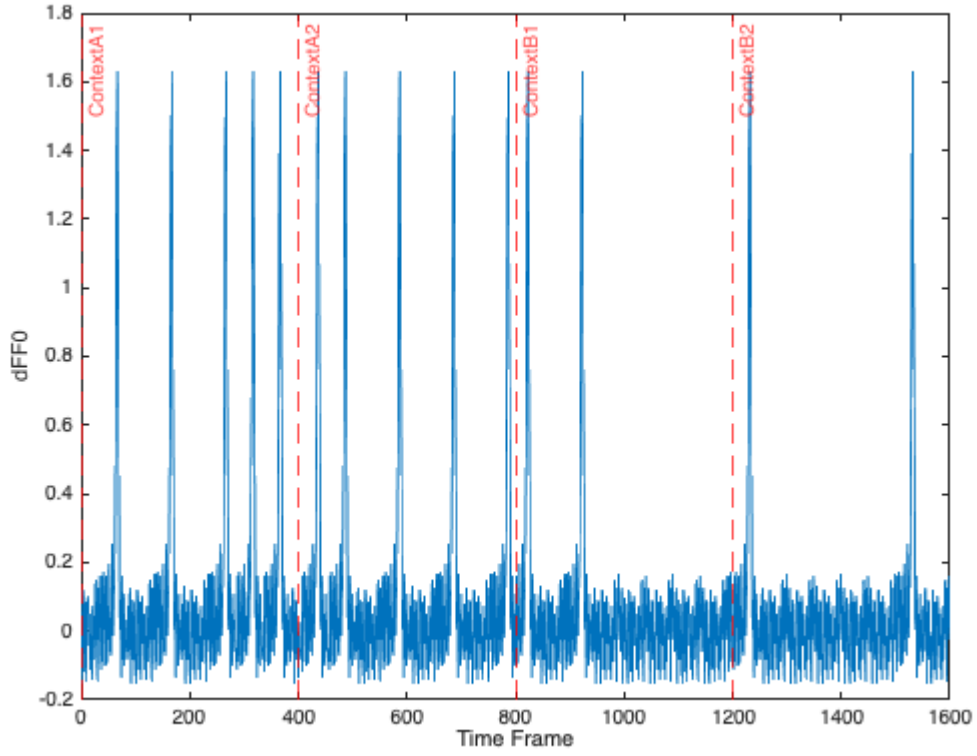

```
% high activity data
high_activity_data = [];
for ff = 1:(num_of_recording_frames_of_a_session/numel(y2))
    r = rand(1);
    if r > 0.3
        y = one_event_trace;
    else
        y = no_event_trace;
    end
    high_activity_data = [high_activity_data, y];
end

% low activity data
low_activity_data = [];
for ff = 1:(num_of_recording_frames_of_a_session/numel(y2))
    r = rand(1);
    if r > 0.8
        y = one_event_trace;
    else
        y = no_event_trace;
    end
    low_activity_data = [low_activity_data, y];
end

figure; t = tiledlayout("flow");
```

```
nexttile
plot(low_activity_data); legend("Low active",Location="northoutside")
nexttile
plot(high_activity_data); legend("High active",Location="northoutside")
ylabel (t, "dFF0");
xlabel(t, "Time Frame");
```




```
f = Frame_idx == "ContextA1";
d = high_activity_data;
for cc = 1:num_of_recording_cells
    r = randperm(num_of_recording_frames_of_a_session,1);
    dFF0_data(f,cc) = [d(r:end),d(1:r-1)];
end

f = Frame_idx == "ContextA2";
d = high_activity_data;
for cc = 1:num_of_recording_cells
    r = randperm(num_of_recording_frames_of_a_session,1);
    dFF0_data(f,cc) = [d(r:end),d(1:r-1)];
end

f = Frame_idx == "ContextB1";
d = low_activity_data;
for cc = 1:num_of_recording_cells
    r = randperm(num_of_recording_frames_of_a_session,1);
    dFF0_data(f,cc) = [d(r:end),d(1:r-1)];
```

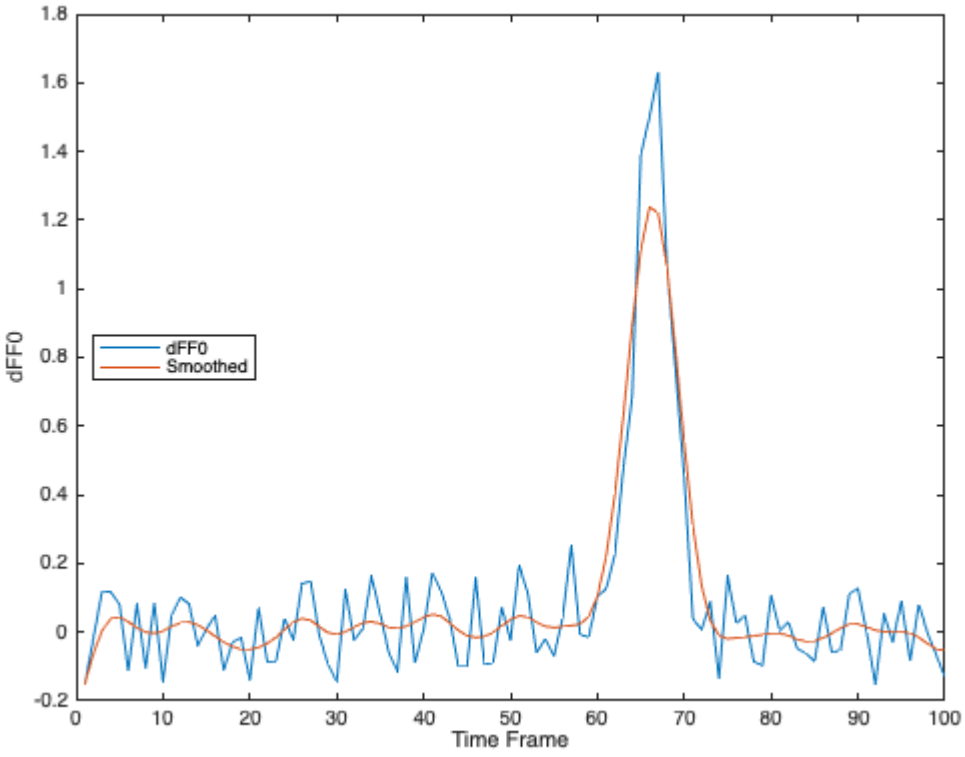
```
end

f = Frame_idx == "ContextB2";
d = low_activity_data;
for cc = 1:num_of_recording_cells
    r = randperm(num_of_recording_frames_of_a_session,1);
    dFF0_data(f,cc) = [d(r:end),d(1:r-1)];
end

figure;
plot(dFF0_data(:,1));
xline(1,'--r',Session_labels(1));
xline(num_of_recording_frames_of_a_session+1,'--r',Session_labels(2));
xline(num_of_recording_frames_of_a_session*2+1,'--r',Session_labels(3));
xline(num_of_recording_frames_of_a_session*3+1,'--r',Session_labels(4));
ylabel ("dFF0");
xlabel("Time Frame");
```



Calcium Event Detetion

1. Smoothed_dFF0_data: 1600 frames-by-30 cells matrix
2. Normalized_dFF0_data: 1600 frames-by-30 cells matrix
3. First_diff_dFF0_data: 1600 frames-by-30 cells matrix
4. Cal_Eve_Binary_data: 1600 frames-by-30 cells matrix

```
z_threshold = 2;

Smoothed_dFF0_data = dFF0_data*0;
[bFilt,aFilt] = butter(2,  2/(recording_Hz/2), 'low');
for ss = 1:numel(Session_labels)
    d1 = dFF0_data(Frame_idx == Session_labels(ss),:);
    smooted_d1 = filtfilt(bFilt,aFilt,d1);
    Smoothed_dFF0_data(Frame_idx == Session_labels(ss),:) = smooted_d1;
end

figure;
plot (dFF0_data(1:100,1)); hold on
plot (Smoothed_dFF0_data(1:100,1)); legend(["dFF0","Smoothed"],Location="west");
ylabel ("dFF0");
xlabel("Time Frame");
```




```
Normalized_dFF0_data = dFF0_data*0;
for ss = 1:numel(Session_labels)
    d1 = Smoothed_dFF0_data(Frame_idx == Session_labels(ss),:);
    d1_sd = std(d1,0,1);
    normalized_d1 = d1./d1_sd;
    Normalized_dFF0_data(Frame_idx == Session_labels(ss),:) = normalized_d1;
end

First_diff_dFF0_data = dFF0_data*0;
```

```
for ss = 1:numel(Session_labels)
    d1 = Normalized_dFF0_data(Frame_idx == Session_labels(ss),:);
    d1_diff = diff(d1);
    zeros_d1_diff = [zeros(1,num_of_recording_cells);d1_diff];
    First_diff_dFF0_data(Frame_idx == Session_labels(ss),:) = zeros_d1_diff;
end

Cal_Eve_Binary_data = dFF0_data*0;
ones_data = ones(size(dFF0_data));
binary_threshold = Normalized_dFF0_data > z_threshold & First_diff_dFF0_data > 0;
Cal_Eve_Binary_data(binary_threshold) = ones_data(binary_threshold);


figure;
plot (Normalized_dFF0_data(1:100,1)); hold on
plot (First_diff_dFF0_data(1:100,1));
yline(z_threshold,'--r',"2SD");
stem(Cal_Eve_Binary_data(1:100,1));
legend(["Normalized","1st diff","2SD","calcium events"],Location="northwest");
ylabel ("Normalized dFF0");
xlabel("Time Frame");
```